THE WATER-RELATIONS OF *BELBA GENICULOSA* OUDMS. AND OTHER SPECIES OF ORIBATID MITES

BY

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INTRODUCTION.

It is often said that insects are the most successful colonizers of diverse land habitats, but quantitative estimation by soil ecologists shows that oribatid mites are equally successful. Most insects, spiders and ticks limit water-loss by a thin, impermeable wax covering which enable some of them to invade dry situations, but a few insects and terrestrial crustaceans lack such a cover and are thus restricted to permanantly damp surroundings.

There is little information on the water-relations of the Oribatidae and what follows compares their water-balance with that of other terrestrial arthropods. This is the first of a series of papers on the physiology and behaviour of such mites.

MATERIAL.

This work deals mainly with a common litter species, *Belba geniculosa* Oudms. (syn. *Damaeus onustus* Koch). Some experiments were also done with other « indicator species » of mites from different environments.

The mites were cultured in small petri dishes on moist plaster-of-Paris and given detritus from their habitat as food. When necessary, they were killed by potassium cyanide or by ammonia vapour.

METHODS.

Batches of recently killed mites were weighed on a sensitive torsion balance (accurate to 0.02 mg), put in small desiccators in dry air for half an hour at different

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constant temperatures and then re-weighed. The micro-desiccators were filter crucibles with sintered discs as bases, of porosity 100-120 μ (manufactured by A. Gallenkemp & Co. Ltd.). The smaller chamber of the crucible (area 26 mm × 2 mm) was covered by a perspex square with capillary tube attachment to allow for air pressure adjustments and sealed with silicone grease. Dry air was obtained with tubes (2.5 cm × 2.5 cm) of phosphorus pentoxide placed inside the larger chamber of the crucible which was then closed with a rubber bung wrapped in polythene (Fig. 1). Cobalt thiocyanate paper (SOLOMON, 1945) placed on to the floor of the cell showed that the required humidity was quickly obtained. The desiccators were weighed down with lead and submerged in a water-bath thermostatically controlled to within 0.1°C of the required temperature. Comparisons of simultaneous thermistor readings of the temperature of the water with that of the air inside the desiccators showed that thermal equilibrium was reached in under 5 minutes.

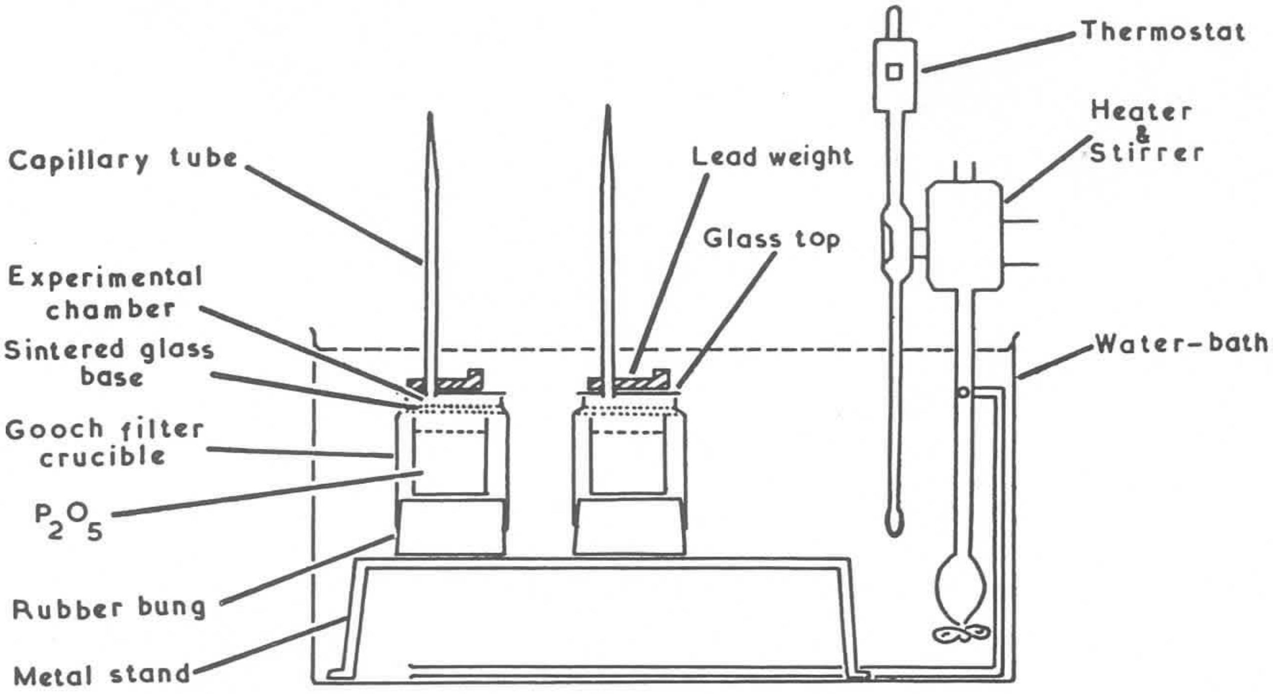

FIG. 1. — Apparatus for investigating the water-relations of oribatid mites.

Because it is difficult to measure the cuticular surface of an arthropod, the rates of evaporation at different temperatures were calculated as the weight of water lost per milligram. However, for comparison with other arthropods, the surface area of the mites was calculated as follows. Mites were placed in lactic acid, squashed under a slide and coverslip and their entire body-surfaces, including the appendages, were traced out on to squared paper using a micrometer eyepiece and strong microscope illumination. From these results, the value of k in the formula $k = \frac{S}{W^{\frac{2}{3}}}$ (WIGGLESWORTH, 1945) was found, where S is the body-surface area in sq mm and W the weight of a mite in mg. Table 1 shows the results obtained, which compare with

Agriotes larva, 11.0 ; *Tenebrio* larva, 8.4 ; *Calliphora* larva, 6.8 (WIGGLESWORTH, 1945) ; *Paradesmus*, 15.0 (CLOUDSLEY-THOMPSON, 1950) ; *Oniscus*, 13.6 ; *Porcellio*, 12.4 ; *Ligia*, 12.3 (EDNEY, 1951) and *Calliphora*, 10.0 ; *Lithobius*, 12.0 (MEAD-BRIGGS, 1956).

TABLE 1.

The surface area/weight relationship of certain species of oribatid mites.

Species	*Micrometer units*	*Surface area*	*Weight*	$W^{\frac{2}{3}}$	$k = \frac{S}{W^{\frac{2}{3}}}$
	(400 : 1.00 sq mm)	(sq mm) (S)	(mg) (W)		
Humerobates rostrolamellatus Grandjean	465	1.12	* 0.097 (49)	0.2110	5.3
Belba geniculosa Oudms	1580	3.95	0.700 (5 × 5)	0.7880	5.0
Platynothrus peltifer (Koch)	240	0.60	0.047 (30)	0.1302	4.6
Euzetes globulus (Nic.)	600	1.50	0.362 (15)	0.5078	2.9
Steganacarus magnus (Nic.)	630	1.57	0.291 (14)	0.4390	3.8
Fuscozetes fuscipes (Koch)	300	0.75	0.078 (40)	0.1825	4.1
Hypochthonius rufulus (Koch)	225	0.56	0.033 (40)	0.1029	5.4
Nanhermannia nana (Nic.)	225	0.64	0.018 (75)	0.0687	9.3

* Calculated from total number of mites in brackets.

EXPERIMENTAL RESULTS.

Preliminary experiments.

a) *Water-loss in still or moving air.*

Evaporation (i.e. water-loss) in arthropods can be measured in still air or in moving air but usually no differences have been shown between them. Similar comparisons were made with adult *B. geniculosa* by keeping them in desiccators in still dry air or in circulating dry air (*c.* 1,000 cc per min.) at 20°C and weighing them 1-3 times a day for 4-5 days, the drying agent (anhydrous silica gel) being changed twice a day.

TABLE 2.

The mean percentage weight loss of *B. geniculosa* in still or moving air.
Temperature 20°C (± 2.3°C).

Replicate	*Mean time weighed (hrs.)*	*Mean weight loss in still air (%)*	*Mean weight loss in moving air (%)*	*Total number of records*	*F-test*
1	42.1	72.0	66.6	7	3.09
2	40.5	68.5	65.2	8	6.31
3	65.3	67.5	63.5	6	2.49

Replicates 1 and 3 not significant at 0.001 level.
Replicate 2 not significant at 0.01 level.

No significant differences in evaporation were obtained (Table 2). After 114 hours the mites lost 69.3 % of their original body-weight in still air and 65.1 % in moving air ; subsequent experiments were therefore made in still air.

b) *Water-loss and time-lapse.*

Different batches of adult mites were exposed to dry air at 20°C, 30°C, 40°C, 45°C, 50°C, 55°C and 60°C and the rates of evaporation found by weighing them every 15 minutes for 90 minutes. Results (Fig. 2) showed that (a) the lower the temperature, the lower the evaporation rate, (b) the longer the exposure in dry air, the greater the evaporation rate, (c) the rate of water-loss is greatest from 0-30 minutes at 45°C and over and (d) there is a sharp increase in water-loss at 40-45°C suggesting a physical change in the water-retaining properties of the integument.

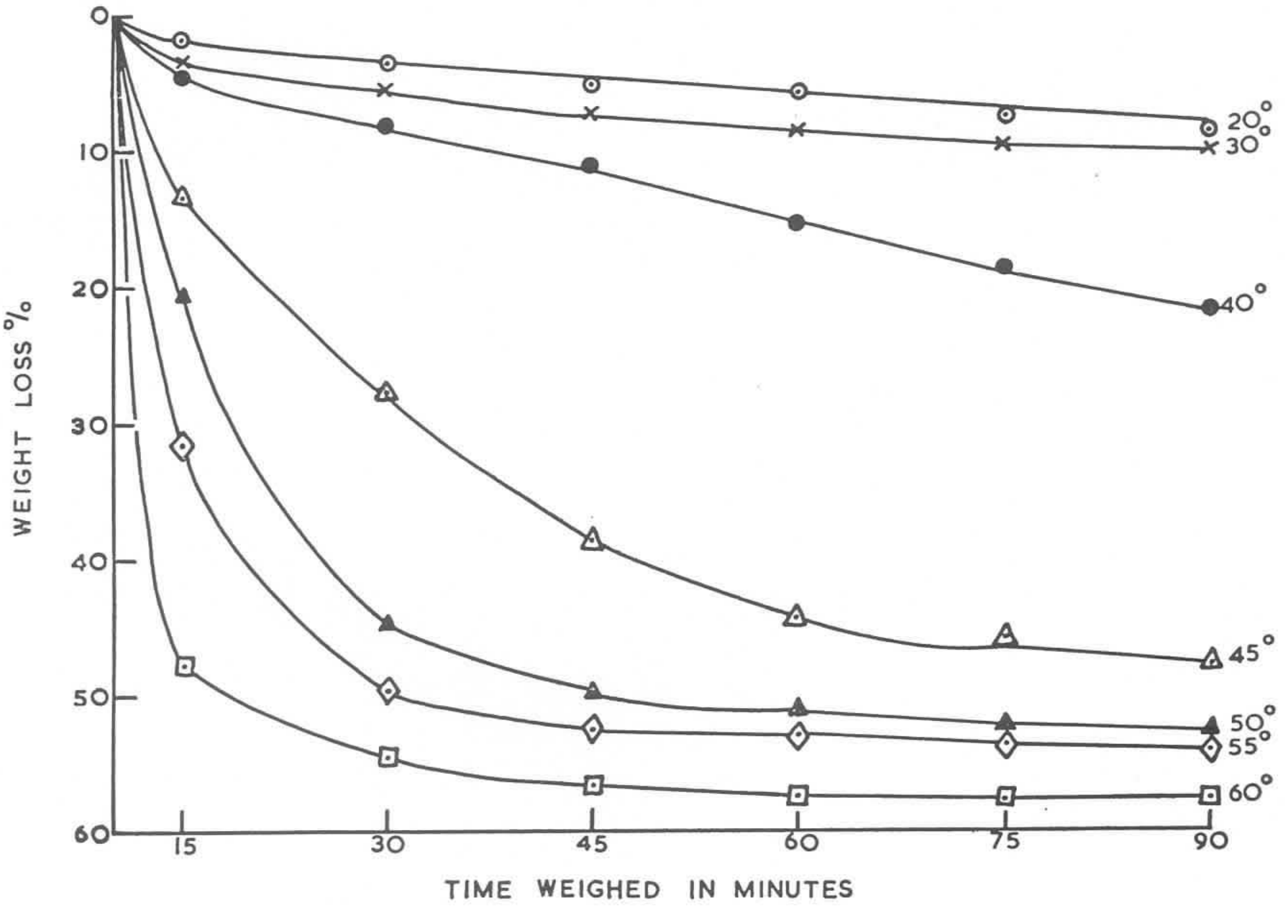

FIG. 2. — The relationship between water-loss and time-lapse with adult *B. geniculosa* at different temperatures in dry air.

1. EVAPORATION AND CUTICLE PERMEABILITY.

a) *Results with B. geniculosa.*

Dead mites were exposed to 0 % R. H. for half an hour at each temperature from 10°C to 70°C and the evaporation rate at these temperatures was then found by loss in weight. There was little evaporation between 10°C-40°C but beetwen 40°C-50°C it greatly increased ; there was little further evaporation between 60°C-70°C. Oven dry weights (105°C for 18 hrs.) showed that most of the original water-content of the mites was lost by the end of the experiment. The curve obtained (Fig. 3, A)

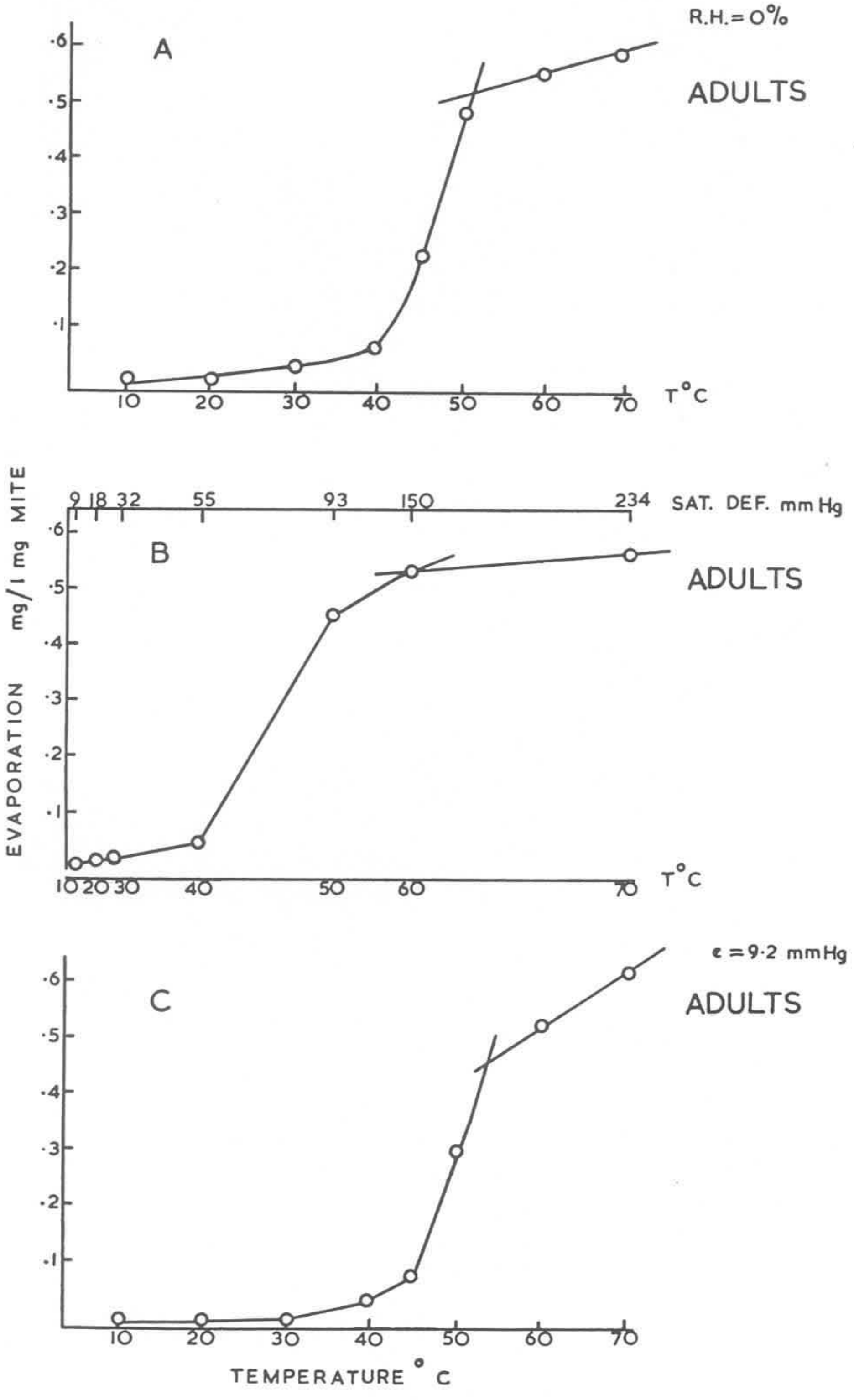

FIG. 3. — The relationship between evaporation and temperature with adult *B. geniculosa*. Further explanation in the text.

is similar to that of many arthropods with a waterproofing layer, the sudden increase in water-loss corresponding to the " critical temperature " (WIGGLESWORTH, 1945) or to the " transitional temperature " (BEAMENT, 1958) of other arthropods [1]. The critical temperature is also apparent when evaporation is related to a saturation deficiency corresponding to 0 % relative humidity (Fig. 3, B). Thus there is no linear relationship between saturation deficiency and evaporation.

Excessive evaporation in dry air was prevented by exposing mites to a constant saturation deficiency of 9.2 mm Hg from 10°C-70°C ; thus evaporation only depended on changes in the outer impermeable surface (MEAD-BRIGGS, 1956). The following relative humidities were used to give 9.2 mm Hg : 0 % (at 10°C), 47.4 % (20°C), 71.2 % (30°C), 83.4 % (40°C), 87.2 % (45°C), 90.0 % (50°C), 93.9 % (60°C) and 96.1 (70°C), these date being calculated from Smithsonion Meteological Tables, 1931 ; Table 85. The results (Fig. 3, C) are similar to those in dry air, with a critical temperature at 40°C-50°C.

There is no second critical temperature above 40°C-50°C, suggesting the lack of a cement layer in the epicuticle (BEAMENT, 1960).

b) *Results with other stages of B. geniculosa.*

The family Belbidae characteristically cover the dorsal region of their body (hysterosoma) with their cast skins, particles of sand and fragments of litter and twigs. The adult integument is strongly sclerotised and is generally smooth, but the juveniles, apart from their appendages, have a thin, pliable, opaque, non-sclerotised and dirt-covered skin (Plates 1 & 2). This behaviour was investigated to show whether it was associated with their water-balance.

Experiments in dry air at different temperatures were done with tritonymphs, deutonymphs and larvae. In some experiments the dirt covering their integument was carefully removed with fine entomological pins mounted on wooden handles, and the mites then left for about 24 hours in moist plaster-of-Paris cells before killing them. Fig. 4 gives the results, showing that evaporation from tritonymphs and deutonymphs was slightly higher with clean mites than with normal (dirt-covered) mites, probably owing to superficial integumental damage when removing the dirt. However, all stages, either exposed or dirt-covered, had a critical temperature of about 40°C. Thus the extraneous integumental material is probably not associated with their water-relations, since excessive evaporation is prevented by a waterproofing layer. Other reasons for this behaviour will be suggested later.

c) *Results with other species of oribatid mites.*

Similar experiments were done with different " indicator species " of oribatids, but a narrower temperature range was sometimes used since evaporation was much more rapid in some species than in others. These mites are smaller than *B. geniculosa* and so larger numbers were used; water-loss is expressed as evaporation in milligrams per mite per half hour, the results being comparable with those of *B. geniculosa*. Loss in weight of a small aluminium foil cup (0.5 cm diam.) filled

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[1] Curves of this kind have been criticised by SHAW (1955), HOLDGATE and SEAL (1956) and EDNEY (1958) because insufficient attention has been paid to the steeply rising drying power of the air, but as WIGGLESWORTH (1957) points out, this still affords the best way of comparing cuticle permability in arthropods.

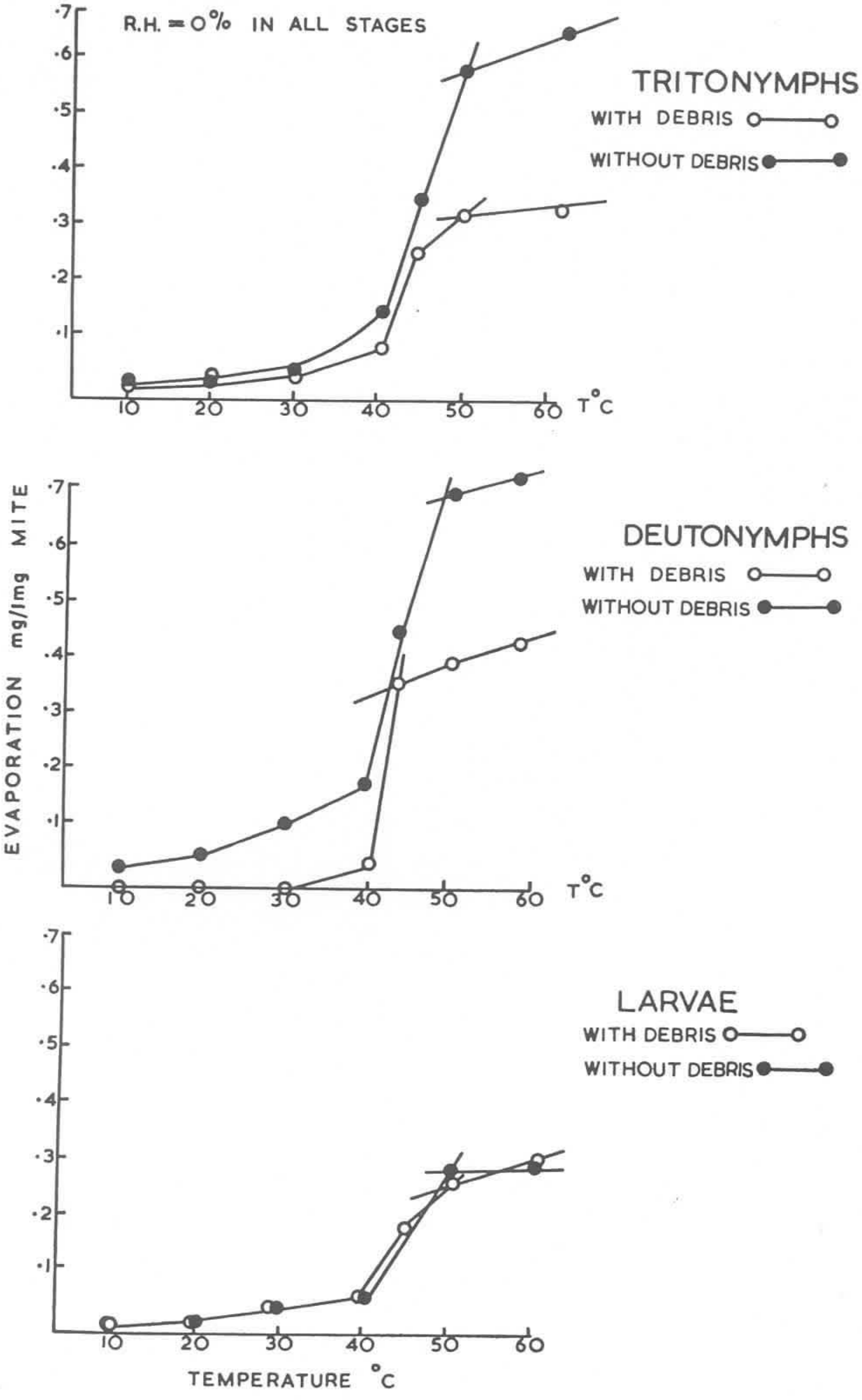

FIG. 4. — The relationship between evaporation and temperature with the juvenile stages of *B. geniculosa*.

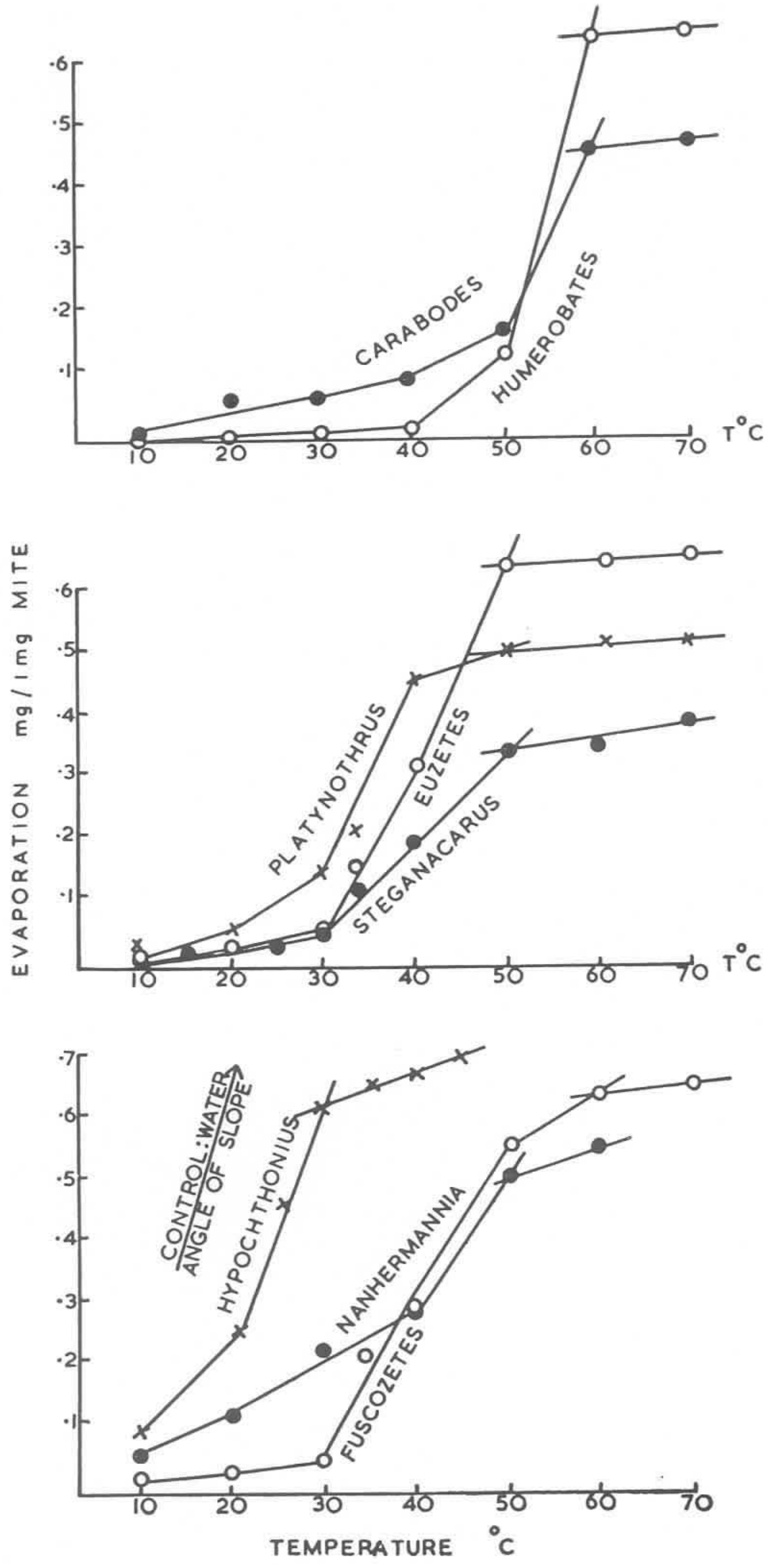

FIG. 5. — The relationship between evaporation and temperature with different species of oribatid mites in dry air. (*Top* : xerophyls ; *middle* : meso-hygrophyls ; *bottom* : hygrophyls).

with water is shown for comparison (control). Comments on these results (Fig. 5) will be given in the discussion.

2. THE EFFECTS OF ABRASIVE AGENTS ON CUTICLE PERMEABILITY.

When inert dusts are brought into contact with insect cuticle there is an increased loss of water, but it is uncertain whether the mechanism is abrasive or adsorptive. *B. geniculosa* was treated with different inert dusts as follows, and attempts were made to determine how the dusts work.

Experimental procedure.

Living or recently killed mites were used. Owing to the prominent setae on the body, the cuticle was abraded with a fine brush repeatedly dipped into the powder. The mites were also dusted with a light jet of activated charcoal to investigate adsorption of the dust ; further details will be given later. After abrasion, any loose particles were removed. The mites were also weighed before and after treatment (no significant differences were noted), and the rate of evaporation was found by leaving them in dry air and re-weighing them at intervals. Untreated mites were used as controls.

a) *Experiments with living and dead mites.*

The evaporation rate of living and recently killed adult *B. geniculosa* in dry air at 20°C ± 1.8°C was found by weighing mites 11 times over a period of 61 hours. More water was lost from dead mites (27.0 % ± 2.9 %) than from living mites (15.3 % ± 2.3 %). Similar results were obtained by LEES (1947) with ticks and by DAVIES and EDNEY (1952) with spiders, but WIGGLESWORTH (1945) found that several species of insects lost water at equal rates whether they were alive or dead.

b) *Experiments with abraded and non-abraded mites.*

Abraded and non-abraded (living and dead) mites were put in desiccators at 0 % R. H. and weighed periodically for 14 hours and then transferred to 100 % R. H. for 48 hours. Untreated living and dead mites were used as controls. Data in Table 3 show that (1) both treated mites and the controls lost weight in 0 % R. H. and partially regained it in 100 % R. H. In saturated air their original weight was not regained. (2) Abraded mites lost more weight in 0 % R. H. and gained more weight in 100 % R. H. than did the controls. (3) After 24 hours in dry air, living abraded and living controls lost weight in the ratio 1.4 : 1.(4) After 48 hours in saturated air, the weight of living abraded mites was four times that of the controls (ratio 3.7 : 1) and the weight of dead abraded mites was one and a half times that of the controls (ratio 1.6 : 1).

TABLE 3.

The effect of abrasion on living and dead *B. geniculosa*. Percentage weight loss over initial weight at 0 % R.H. shown. Untreated mites were used as controls. Temperature 20°C (± 1,9°C).

Time weighed (hrs.)	*Abraded alive*	*Control alive*	*Abraded dead*	*Control dead*
6	19.6	8.3	11.1	10.5
18	37.5	19.6	27.3	24.3
24	40.6	27.4	32.7	31.8
	Percentage weight regained in 100 % R.H.			
48	10.6	4.5	4.9	3.9
72	19.0	5.1	8.6	5.6

Living abraded mites are thus probably capable of actively absorbing water from staurated air through the surface of their damaged integument, since they gained twice as much weight (1.9 : 1) as dead abraded mites. Dead abraded mites were unable to do so, water vapour being taken in passively. Living desiccated abraded mites rapidly increased in weight in saturated air. LEES (1947) showed

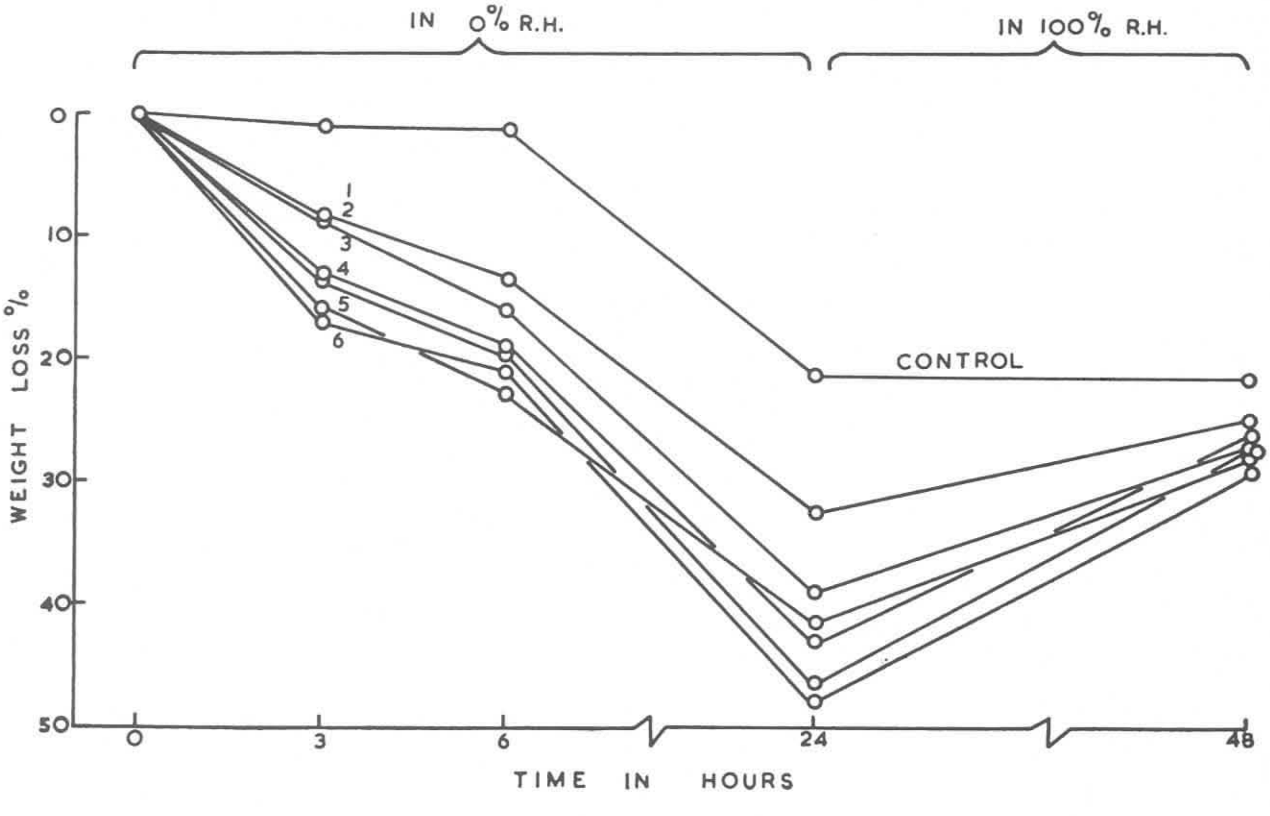

FIG. 6. — The effect of various abrasive dusts on evaporation with *B. geniculosa* at about 20°C. The dusts used were (1) aluminium hydroxide powder B.T.C., (2) activated charcoal, (3) silicon carbide No. 220, (4) silicon carbide No. 440, (5) alumina white No. 2 and (6) activated alumina type « H ».

that in ticks there was a delay of 24 hours before they increased in weight under similar conditions. Thus, the epidermal activity of ticks was temporarily delayed whereas that of mites began soon after abrasion. The living mites all died in three days ; their weights in 0 % or 100 % R. H. thereafter closely followed that of initially dead mites, thus providing further evidence that living mites have a different water-balance to dead mites.

c) *The effect of different abrasive dusts.*

Groups of mites were treated with six abrasives which differed widely in physical structure and properties, and then put in 0 % R. H. and re-weighed after 3, 6 and 24 hours. The results are given in Fig. 6, showing the rate of evaporation was greater with treated mites than with untreated controls, the rate of evaporation depending on the abrasive used. Activated alumina dust was the most effective and aluminium hydroxide powder the least effective. When left in 100 % R. H., all the mites, except the controls, increased in weight (Fig. 6). But unlike results obtained by GLYNNE-JONES (1955), hard, " gritty " abrasives were more effective on mite cuticle than soft amorphous powders. Table 4 gives the relationship between the efficiency of the abrasives and their physical structure, showing that the more " gritty " abrasives wirth sharp angular sides damaged the integument more effectively than the soft amorphous powders.

TABLE 4.

The physical characteristics of various abrasive dusts and the rate of water-loss of treated *B. geniculosa* for the first 3 hours over the controls. Temperature *c.*20°C.

Abrasive dust	*Particle size* (mm)	*Description*	*Evaporation rate* (%) *overs controls* 0-3 hrs.
Aluminium hydroxide powder B.T.C.	0.001 or less	Soft powdery aggregates	6.7
Activated charcoal (fine)	0.001–0.003	Ditto	7.2
Silicon carbide No. 220	0.015–0.15	Gritty, with sharp irregular sides	11.5
Silicon carbide No. 440	0.003–0.01	Powdery, with sharp irregular edges	12.0
Alumina white No. 2	0.004–0.02	Powdery, with spherical aggregates	14.6
Activated alumina type " H "	0.004–0.01	Very brittle and gritty, with sharp edges	15.4

d) *The effectiveness of abrasion and adsorption.*

It is uncertain how inert dusts work on the surface of arthropod cuticles, except that they are abrasive or adsorptive. BEAMENT (1960) showed that the critical temperature persisted with both treatments. The abrasive properties of charcoal powder were found by stroking dead mites 20 or 40 times with a soft brush dipped into the abrasive and then determining the evaporation rates from 20°C to 60°C. The results (Fig. 7) proved that charcoal acts as an abrasive, giving greater efficiency with the longer treatment and a less well-defined critical temperature than in the untreated controls.

The adsorptive action of the same powder was found as follows. Living or recen-

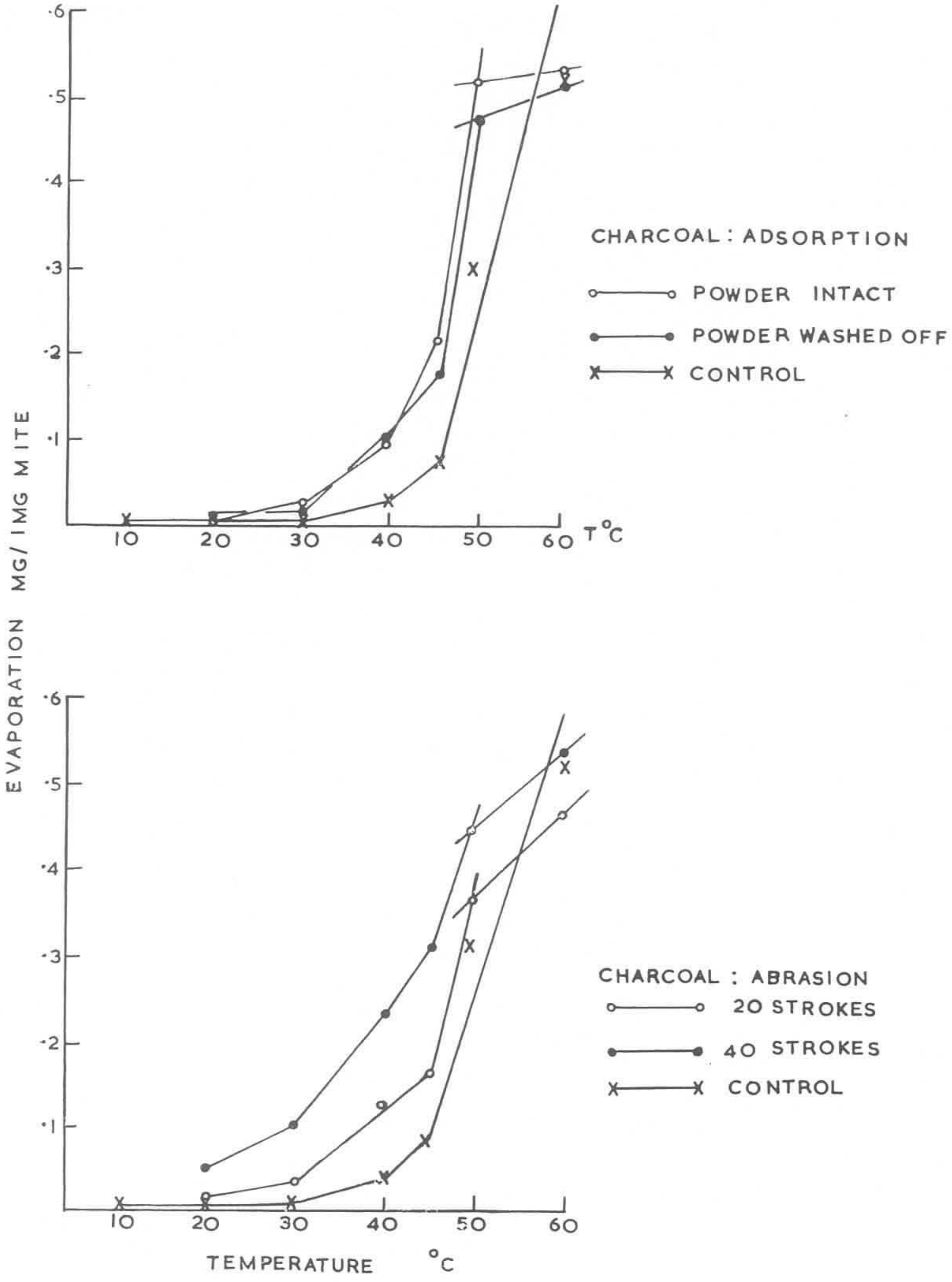

FIG. 7. — The effect of adsorption and abrasion with *B. geniculosa*.

tly killed mites were placed in a slanting capillary tube (0.8 cm × 20 cm) open at both ends, and a fine uniform jet of activated charcoal powder was blown over them with an air-pump joined to a fine glass capillary nozzle. After one minute half the living mites were removed and killed ; the remainder were repeatedly dipped into distilled water to remove the superficial dust, blotted to remove excess surface water and then killed. The dead mites received the same treatment. The evaporation rates in dry air were then found as usual. The results (Fig. 7) showed that there was no significant differences between mites with or without a powder deposit (washed or unwashed). The dust was as effective with short abrasive treatment as with adsorption treatment, but the evaporation rate was greater with longer abrasion.

3. THE RECOVERY OF IMPERMEABILITY.

WIGGLESWORTH (1945) and LEES (1947) showed that live abraded insects and ticks gradually repaired their integument, and LEES (*loc. cit.*) noted the re-deposition of the wax layer in saturated air. Recovery in both cases was incomplete. Living *B. geniculosa* were abraded with alumina dust and then cultured at 15°C with litter debris as food for 6, 18, 24, 48, 72 and 96 hours. They were afterwards killed and their evaporation rates in dry air found from 20°C-60°C (Fig. 8). The mites gradually replaced their waterproofing cover over 5 days but comparisons with untreated controls suggest that this was incomplete. The initial rates of evaporation at 20°C were as follows : .085 ±.01 (6 hours recovery period) ; .066 ±.01 (18 hrs.) ; .033 ±.01 (24 hrs) ; .024 ± .01 (48 hrs) ; .020 ± .00 (72 hrs) ; .010 ± .00 (96 hrs) and .012 ± .00 (control) per mg mite per half hour. The shapes of the curves show that the longer the recovery period, the sharper the critical temperature and therefore the greater the impermeability. Inconclusive results were obtained by direct microscope observations, and by the polyphenol ammoniacal silver nitrate test.

4. THE EFFECT OF CHLOROFORM ON CUTICLE PERMEABILITY.

Abrasives gave no information on the nature of the cuticle surface. Mites were therefore treated with a wax solvent to determine whether a wax or lipid cover existed.

Dead mites were weighed, put into tubes containing 20 cc of either cold (20°C) or hot (45-55°C) chloroform and the containers mechanically shaken for a definite period. The mites were afterwards put into tubes plugged with damp cotton wool for about 30 minutes to allow the chloroform to evaporate in saturated air. The evaporation rates were then found in dry air from 10°C-70°C. In all treatments (Fig. 9) there was considerable loss of water but in no instance, except for the controls, is there a marked indication of a critical temperature. Hot chloroform (results not shown) is much more effective, resulting in significantly higher evaporation rates, but this may also be due to loss of body-fats. The shapes of the graphs from curvilinear with long treatment to linear with short treatment suggest that the wax surface is progressively lost the longer the treatment. The results show that the waterproof layer of mite integument is of a waxy or lipid material, soluble in chloroform and probably similar to insect epicuticular wax.

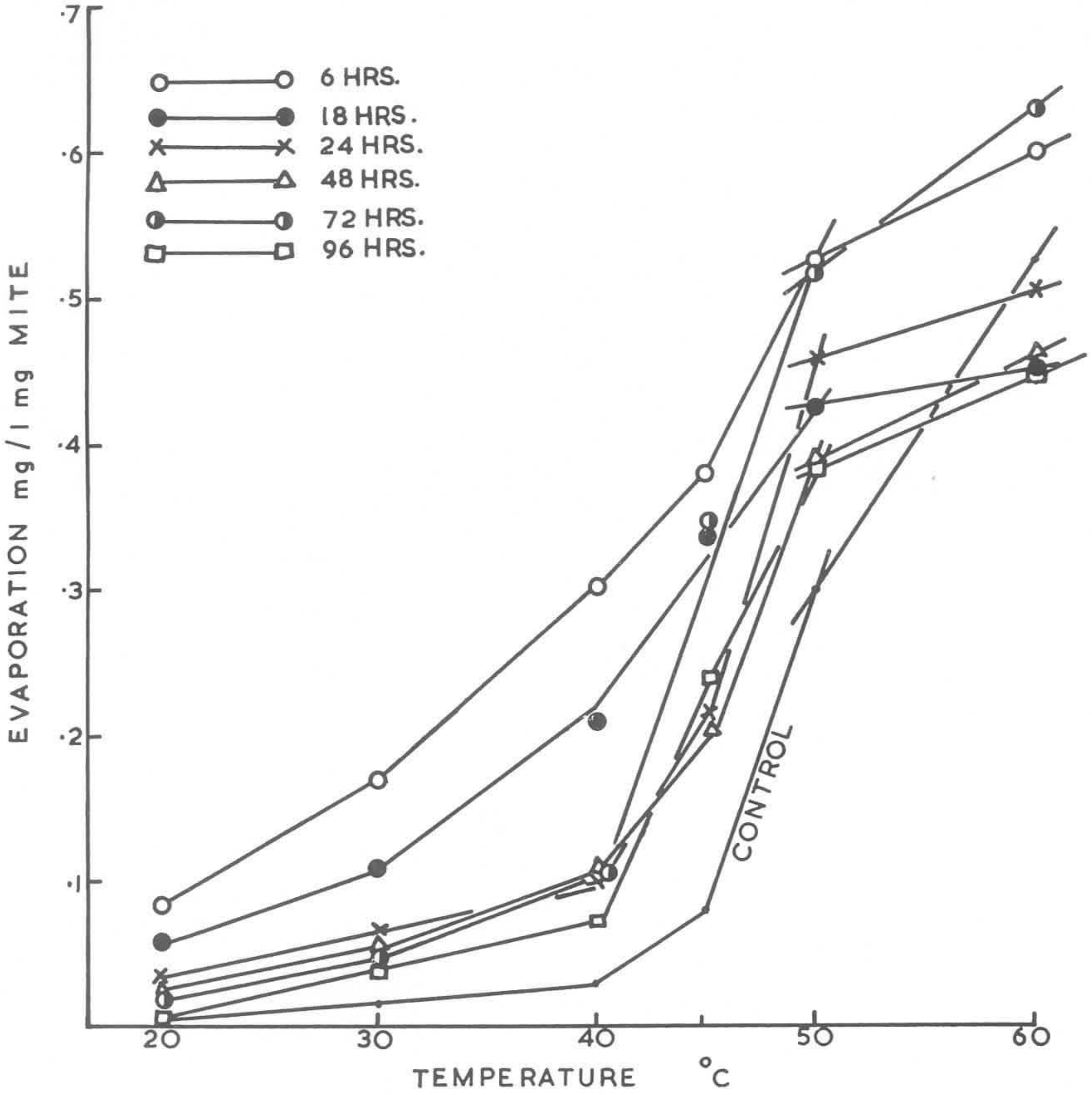

Fig. 8. — The recovery of impermeability with adult *B. geniculosa*. The mites were abraded and allowed to recover in saturated air at 15°C at times shown.

5. The site of water-loss.

a) *The anatomy of the respiratory system of B. geniculosa.*

Like most terrestrial arthropods, the Acarina usually have a tracheal system for respiration. It is impossible to summarise the diversity of such respiratory systems amongst oribatid mites (see e.g. GRANDJEAN, 1933 and 1934), but broadly speaking, they may have well-developed tracheae with three pairs of podosomatic stigmata (Aptyctima) or tracheae may be absent (Ptyctima). NICOLET'S (1855) account of the tracheal system of *B. geniculosa* was criticised by MICHAEL (1885). MICHAEL (*loc. cit.*) found four pairs of tracheae, each opening into the fused coxal regions (acetabula) of legs I to IV, the first two pairs being double and the remaining ones single. However, GRANDJEAN (1934) described only three pairs of tracheae, the last pair, opening into leg IV being absent. Attempts were made to clear the existing confusion by dissecting this species under a binocular microscope. Fig. 10 shows a general semi-diagrammatic view of the tracheal system of *B. geniculosa*. The first pair of tracheae is finer than the others but is clearly double with a short common trachea opening on the dorsal acetabular region of leg I. The second pair, also double, join a short common bulbous opening which opens in turn into an oval air-filled slit at the base of a chitinous ridge (apodème séjugal ; GRANDJEAN, 1934) separating legs II and III. However, the site of this external opening, unlike the others, is difficult to locate as the whole area is strongly sclerotised. The third

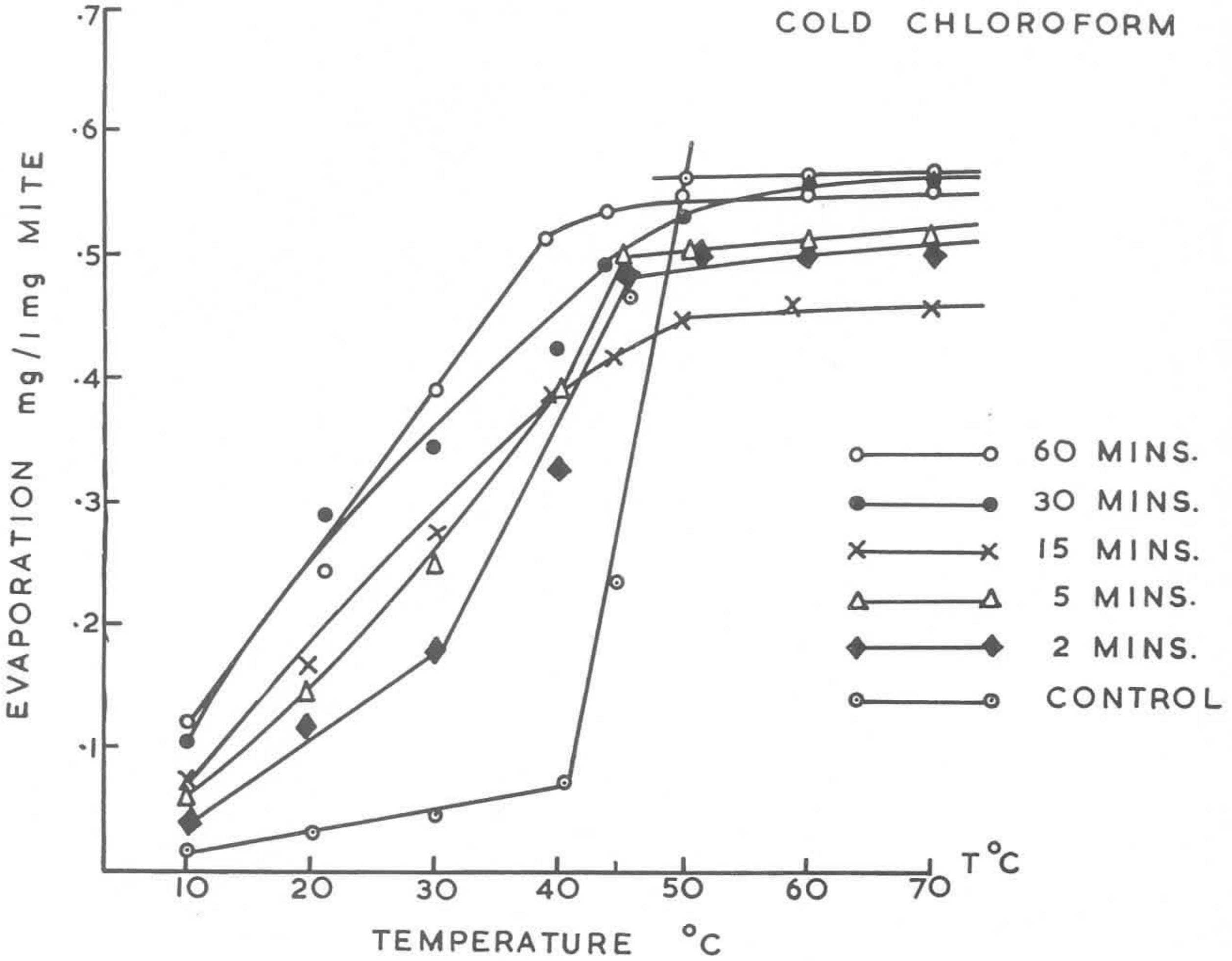

FIG. 9. — The effect of chloroform treatment on evaporation in dry air at different temperatures with *B. geniculosa*. The mites were left in the chloroform at 20°C for times shown.

trachea is single and opens into the mid-posterior region of the acetabulum of leg III. It loops across the fourth acetabulum and then passes backwards but otherwise has no further connection with it. These observations generally agree with those of GRANDJEAN'S (1934). No spiracular closing mechanisms were found ; attempts to inject the tracheae with cobalt sulphide (WIGGLESWORTH, 1950) or immersing the animals in a tracheal stain (PANTIN, 1959) were unsuccessful. No other observations were made on other possible respiratory sites, namely the pit-like depressions on the body (" porose areas "), the air-filled cavities of the legs (GRANDJEAN, 1940) and the Organ of Claparède (GRANDJEAN, 1946 and 1955).

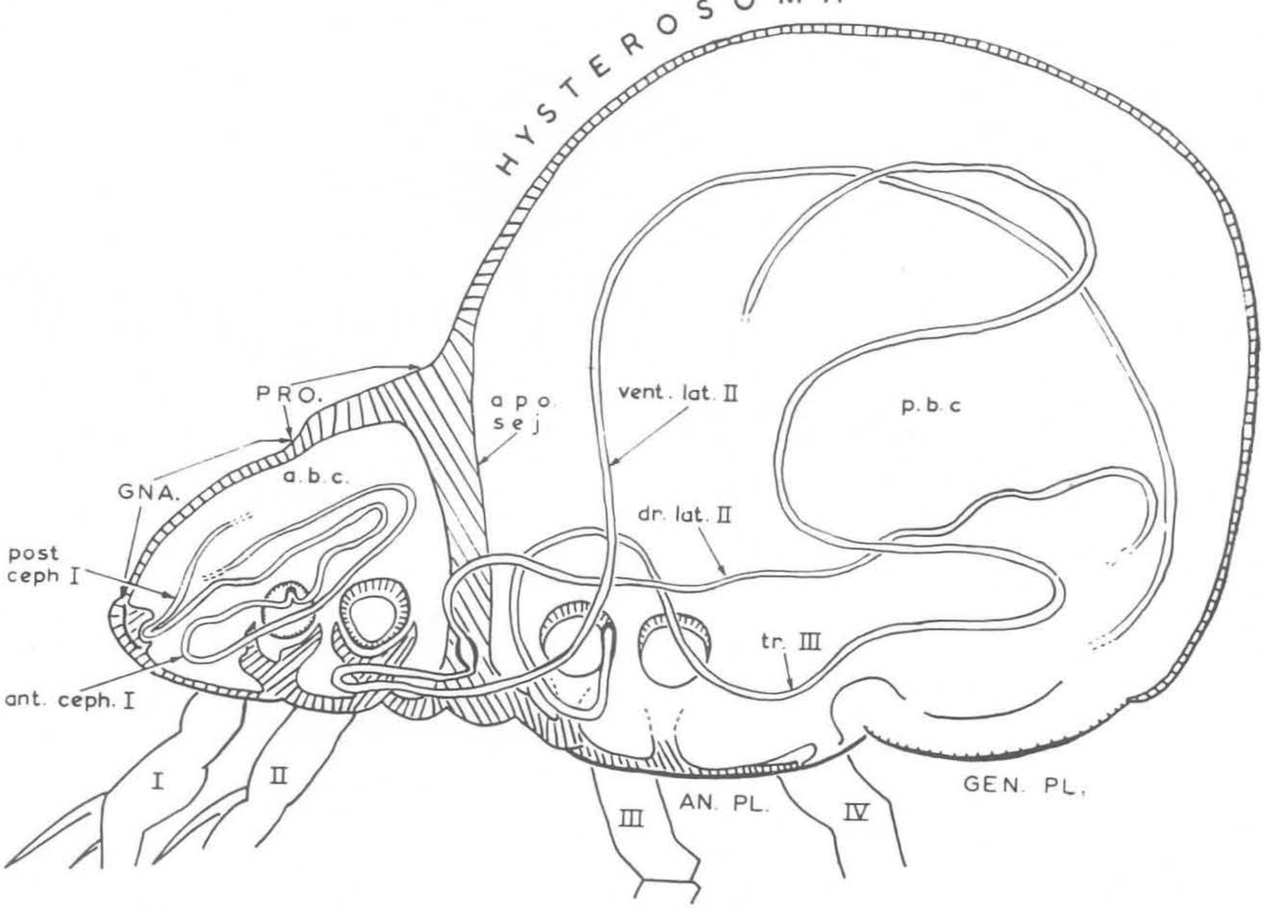

FIG. 10. — Semi-diagrammatic view of the tracheal system of *B. geniculosa* (adult). Longitudinal section. Length of mite : about 1.2 mm. Tracheal endings not shown.

Legend

a.b.c., anterior body-cavity. — p.b.c., posterior body-cavity. — GNA, gnathosoma. — PRO, propodosoma. — apo. sej., apodème séjugal (i.e. chitinous ridge separating the anterior and posterior body-cavity). — AN. PL., anal plates. — GEN. PL., genital plates. — ***Trachea I***, anterior and posterior cephalic tracheae opening into the dorsal region of the acetabulum of leg I. — *Trachea II*, dorsal and ventral tracheae opening at the base (?) of the apodème séjugal. — *Trachea III*, single trachea opening into the dorso-posterior region of the acetabulum of leg III.

b) *The effect of carbon dioxide on water-loss from the tracheae.*

(1) *Dead mites.* Since it was impossible te block the spiracles owing to their inaccessibility, water-loss from the tracheae was found by another method. When insects (MELLANBY, 1934), ticks (LEES, 1946) and spiders (DAVIES and EDNEY, 1952) are gassed with carbon dioxide, their spiracles are left open and they thus lose more water than untreated controls. Similar experiments were done with *B. geniculosa* at 19°C (± 2.3°C) by axphyxiating them for 10 minutes in a slow-moving stream of water-saturated air containing 5 % carbon dioxide. They were then killed, put in desiccators in dry air and weighed at least 3-4 times daily for 4 days. Water-loss from untreated dead mites were used as controls. Table 5 shows that there was no statistical difference between the CO_2 treated mites and the controls (P = 0.05-0.01). It is therefore concluded that either a negligable amount is lost from the spiracles (assuming that they were open in treated mites and closed in the controls) or that this loss is the same whether they are open or closed.

TABLE 5.

Mean percentage weight loss (expressed as % original weight) of dead *B. geniculosa* after 4 days in dry air, with and without 5 % CO_2. Temperature 19°C (± 2.3° C).

Replicate	*Total number of readings*	*Mean time in hours*	*Air + CO_2*	*Air only (control)*	*t-test*
1	16	66.7	42.0	39.8	0.05–0.01
2	11	74.3	54.6	53.3	0.001

Grand mean : Hours 70.5 ; Air + CO_2 57.4 ; Air only 58.6.

(2) *Living mites.* Adults were put in small glass tubes sealed with fine nylon gauze and suspended in a closed container lined with filter paper moistened with aqueous sulphuric acid to give 98 % R. H. The mites were then weighed daily for 10 consecutive days. A 5 % stream of pure CO_2 was blown through the apparatus every second day and the apparatus then immediately sealed. An average of 4.5 faecal pellets was voided per mite per day, but loss in weight owing to defaecation was ignored. In another experiment, mites in dry air were treated every 3 hours daily for two days with air and air + CO_2 alternately and left in saturated air overnight. No faecal pellets were voided in dry air. Data in Table 6 show that there were no significant differences in evaporation rates between mites in dry air and those in air + CO_2, whether the air was dry or saturated (P = 0.05-0.01). Thus, unlike many insects which have spiracular closing mechanisms, it appears that *B. geniculosa* has no such mechanism. Water-loss from this site is the same whether the mites are alive or dead.

TABLE 6.

The effect of 5 % CO_2 on water-loss with living *B. geniculosa*. Mean percentage weight loss and standard deviation of mean shown. Four batches of 4 adult mites per test, at 20°C (± 2.5°C). Result not significant at 0.05–0.01 level

0 % R.H. (15 hrs.)		*98 % R.H. (10 days)*	
Air	*Air + CO_2*	*Air*	*Air + CO_2*
92.9+1.6	90.9+1.9	92.6+1.8	91.9+1.5

c) *The respiratory system of other oribatid mites.*

These have widely differing respiratory systems. Tracheae are absent in *Hypochthonius rufulus* (Koch) but respiration probably occurs through a porous cuticle (GRANDJEAN, 1934). There is no tracheal system in *Platynothrus peltifer* (Koch) but narrow air spaces beneath the pseudostigmatic organs may be connected with respiratory exchange (GRANDJEAN, 1934 ; TARMAN, 1959). The respiratory system of *Steganacarus magnus* (Nic.) consists of three short pairs of tracheae at the base of the pseudostigmatic hairs (CLAPARÈDE, 1868 ; GRANDJEAN, 1934). GRANDJEAN (*loc. cit.*) has suggested that cuticular respiration probably occurs in this species, but as it covered by a waxy epicuticle this is unlikely to be so. *Humerobates rostrolamellatus* Grandjean has the " normal " tracheal system of more highly developed oribatids such as *Belba geniculosa* Oudms. (GRANDJEAN, 1933). No information is available on the respiratory organs of *Euzetes globulus* (Nic.), *Fuscozetes fuscipes* (Koch) and *Nanhermannia nana* (Nic.), although my observations suggest that the first two species have a tracheal system. *N. nana* has a cuticular pattern marked with small semi-transparant areas, suggesting that these may be the openings of large ducts ; no definite tracheae have been found.

6. LOSS OF WATER BY EXCRETION.

Very little work has been done on the excretory organs of oribatid mites and what follows is a summary of known facts. MICHAEL (1884) criticized NICOLET'S (1855) description of the air sacs found in certain mites, and showed that they were liquid-filled structures which he called super coxal glands. These, with the expulsory vesicles, were stated to act as excretory organs. LEES (1946) found that similar coxal glands in ticks acted as osmoregulators. HUGHES (1959) states that oribatid mites sometimes have paired excretory tubules opening into the alimentary canal at the junction of the colon and the post-colon, but these tubules have lost their function, which has been taken over by the colon. Solid faecel pellets are found in the colon and most of the water is re-absorbed in the post-colon. Guanine granules have been seen in the body-tissues.

Experiments were done to find (a) the average number of faecal pellets and

(b) the water-content of the faecal matter of *Steganacarus magnus*, this species being easily reared in culture. Mites were cultured at 10°C and 20°C in plaster-of-Paris cells moistened with distilled water and fed with discs (0.8 cm diam.) of either recently fallen oak leaves (" fresh " leaves) or decaying leaves (" old " leaves). The number of faecal pellets were counted weekly and the fresh weight, oven-dry weight (105°C for 24 hours) and ash weight (700°C for 6-8 hours) was determined (Table 7). At these temperatures, mites fed on fresh and old leaves voided faecal pellets in the ratio 1 : 1.25 per week respectively ; an average of 5.5 or 9.5 pelets were voided per week at 10°C and 20°C respectively. The water-content of 4,176 faecal pellets was 3.1 % of their fresh weight ; the organic matter contributed 88.5 % and the ash content 8.4 % of their dry weight. Most of the faecal pellets thus consist of undigested (organic) remains, with a very low water-content. Therefore little water is lost by excretion.

TABLE 7.

Faecal production in *Steganacarus magnus* (Nic.) when given freshly fallen or old oak leaf discs (mean results of 2 batches of 25 mites for each experiment shown).

Temperature °C		10		20	
Food material		*Fresh leaf discs*	*Old leaf discs*	*Fresh leaf discs*	*Old leaf discs*
Week	1	169	343	467	708
	2	228	296	548	696
	3	291	312	439	298
	4	333	365	169	331
Total		1,021	1,316	1,623	2,033
Number of pellets per week per mite		5.01	6.06	8.01	10.10

Total number of faecal pellets weighed	: 4,176
Fresh weight 4,176 faecal pellets	: 7.48 mg
Oven-dry weight faecal pellets	: 7.24 mg
Ash weight faecal pellets	: 0.85 mg

7. WATER-LOSS FROM OTHER SITES.

Attempts were made to locate water-loss from other regions of the body by occlusion experiments with shellac varnish or cellulose paint. The following areas were sealed : (a) mouth, (b) anal and genital plates, (c) mouth, anal and genital plates and (d) the legs. Untreated mites were used as controls. The mites were then killed and the evaporation rates found in dry air at 19°C (± 1.7°C) for 4 days. However, the results were so variable that no details are given. Generally, mites with occluded areas (a) or (b) lost weight at approximately the same rates. When all three apertures were sealed simultaneously (c), evaporation was decreased. Water-loss by mites with occluded leg appendages (d) and the controls was similar. The results suggest that evaporation occurs mainly through the mouth, genital and anal apertures.

TABLE 8.

The relationship between certain species of oribatid mites and various physiological and ecological factors.

Species	*Critical temperature* (°C)	*Respiratory surface*	*Dominant type of habitat*	*Conditions in habitat*
—	—	—	—	—
H. rostrolamellatus	60	Tracheal system	Bark of trees	Marked diurnal variations in temperature and humidity
C. labyrinthicus	50	?	Heathland	Ditto
B. geniculosa	40–50	Tracheal system	Litter	Variable but probably fairly stable
E. globulus	35	Probably tracheal system	Litter & sphagnum	Ditto
S. magnus	30–35	Reduced tracheal system	Litter & sphagnum	Ditto
P. peltifer	30–35	Reduced to air-space under pseudostigmata	Sphagnum mostly, sometimes litter	More stable
F. fuscipes	30-35	? but probably tracheal system	Sphagnum	Humidity always saturated, small variations in temperature
N. nana	?absent	Integument and ? tracheae	Sphagnum	Ditto
H. rufulus	Absent	Integument only	Sphagnum	Ditto

8. DISCUSSION, WITH SPECIAL REFERENCE TO HABITAT.

The wax epicuticle is an important feature of successful land arthropods. This results in water-retention, allowing them to occupy even the driest habitats. Most of the mites studied are protected from excessive desiccation by such a cover. The melting point of the wax varies according to the species, and a possible correlation exists between this factor and their environment (Table 8 and Fig. 5). The bigger the ambient temperature and relative humidity range, the higher the melting point of the wax. Thus, it is 50°C to 60°C in two xerophyl species, *C. labyrinthicus* and *H. rostrolamellatus* ; in a " true " mesophyl, *B. geniculosa*, it is 40°C-50°C ; in most meso-hygrophyls it varies from 30°C to 35°C, and there is no wax cover in two hygrophyls, *H. rufulus* and *N. nana*. Lees (1947) found a parallel relationship in ticks.

There is also a broad relationship between the type of respiratory surface and the kind of habitat in which the mites were collected (Table 8). The greater the diurnal temperature and relative humidity changes in the habitat, the greater the specialization of the respiratory surface. Thus, *H. rufulus*, a mite with primitive morphological characters, is only found in permanently damp places (sphagnum) with a stable temperature ; respiration occurs through a porous, non-waxy integument. *N. nana* is also common in such environments and gas exchanges are probably affected in the same way. *P. peltifer* is often collected in sphagnum and more rarely in drier habitats ; the respiratory system is restricted to an air space under the pseudostigmatic organs. *S. magnus*, a widespread species equally distributed in sphagnum and litter, has one reduced to three small pairs of tracheae in the head region. Finally, *B. geniculosa* and *H. rostrolamellatus* have the usual well-developed tracheal system of more advanced mites, and live in climatically variable habitats of loose litter and bark of trees respectively. These relationships are tentative rather than conclusive, for too little is known about the general biology of these mites and the microclimates in which they live.

Under natural conditions, the juvenile stages of *B. geniculosa* cover themselves with their exuviae and dirt particles ; the adults usually do not. This behaviour is not associated with the water-balance of the juveniles. Their colourless, soft skin has a waterproofing cover which is just as effective in preventing water-loss as that of the strongly sclerotized and impermeable integument of the adults. Two possible reasons for this behaviour are suggested. (1) As a protection against predators. Mesostigmatid (predatory) mites are commonly found in similar habitats and presumably the juvenile oribatids are open to attack by these, and other predators. The advantage gained by a protective coat of extraneous matter covering vulnerable exposed areas during the precocious stages of their life-history is obvious ; the adults are well-armoured by a thick skin and hence need little protection. Associated with this behaviour is one of camouflage : the dark particles covering the pale integument of the juveniles follow the colour pattern of their environment and they are thus made inconspicuous. (2) As a protection against excessive damage to the waxy epicuticle. Since the mites are active in the humus and bottom litter layers, the skin surface of this species is exposed to constant mechanical abrasion ; a cover of extraneous material thus acts as a barrier against superficial damage. But as they can quickly repair their damaged waxy layer, this feature may be less important than the first.

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SUMMARY.

1. A study has been made of the water-relations of free-living soil mites (Oribatidae) and especially of a common litter species, *Belba geniculosa* Oudms.

2. There is no significant difference in evaporation in either still or moving dry air.

3. When exposed to a range of different temperatures (10°C–70°C) in either dry air or in a constant saturation deficit of 9.2 mm Hg, the waterproofing cover of the adults has a marked critical temperature of about 45°C ; in juveniles the corresponding temperature is about 40°C.

4. The normal behaviour of covering their soft unsclerotized integument with their cast skins and debris is not associated with the water-relations of the juveniles. This habit may be associated with (a) protection from predators and (b) protection of the waxy epicutile from abrasion.

5. The rate of evaporation of dead mites in dry air is greater than that of living mites.

6. Evaporation in dry air was increased when the integument was abraded. These mites also showed a greater increase in weight in saturated air than did untreated controls.

7. The rate of water-loss of abraded mites depends on (a) the shape but not necessarily the size of the abrasives (soft powders were least effective and hard " gritty " dusts most efficient) and (b) the frequency of the treatment

8. A soft powder acted equally well as either an abrasive or as an adsorptive agent.

9. Living abraded mites slowly recovered their impermeability in saturated air, but this process was not entirely successful.

10. Cold or hot chloroform treatment resulted in a marked increase in evaporation ; the longer the treatment the greater the water-loss. The outer cover is thus a wax.

11. Carbon dioxide has no effect on evaporation with living mites in dry or saturated air, nor is water-loss increased when they are pre-treated with the gas and then killed. *B. geniculosa* has no spiracular closing mechanism.

12. *B. geniculosa* has a well-developed respiratory system consisting of three pairs of tracheae. Comments are given on the observations of other workers.

13. Water-loss probably occurs mainly through the oral, anal and genital apertures. Losses by excretion are negligable.

14. Comments are given on the adaptation of certain species of oribatid mites to different environments on the basis of (a) the critical temperature of their waterproofing cover and (b) their type of respiratory system.

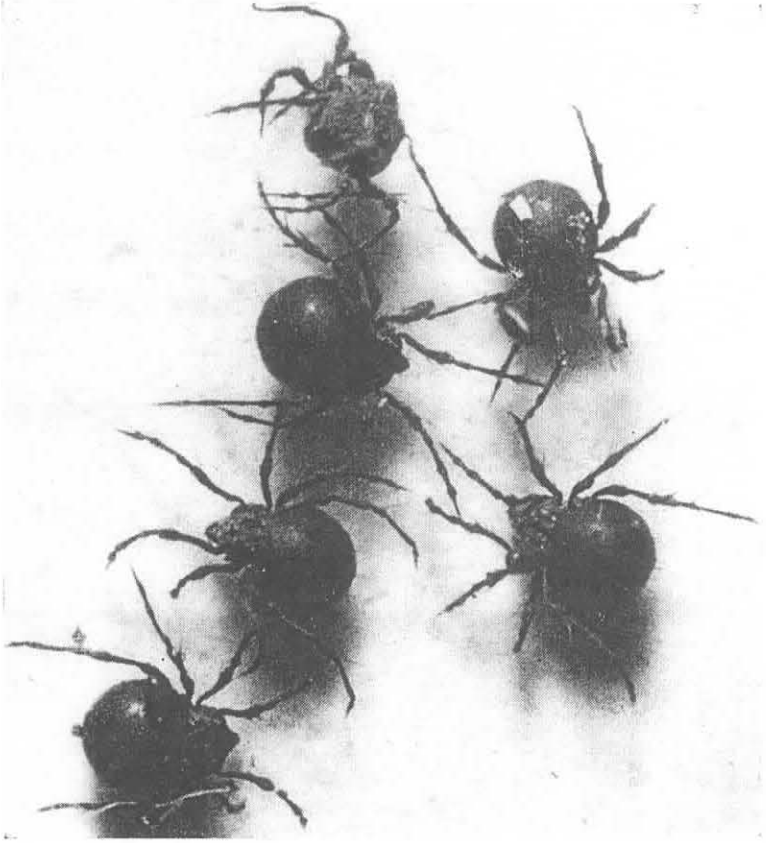

PLATE I.

Smooth-skinned, strongly sclerotized adult *B. geniculosa*. One debris-covered, non-sclerotized tritonymph is seen at the top. Note eggs attached to legs and body of three of the adults (× 10).

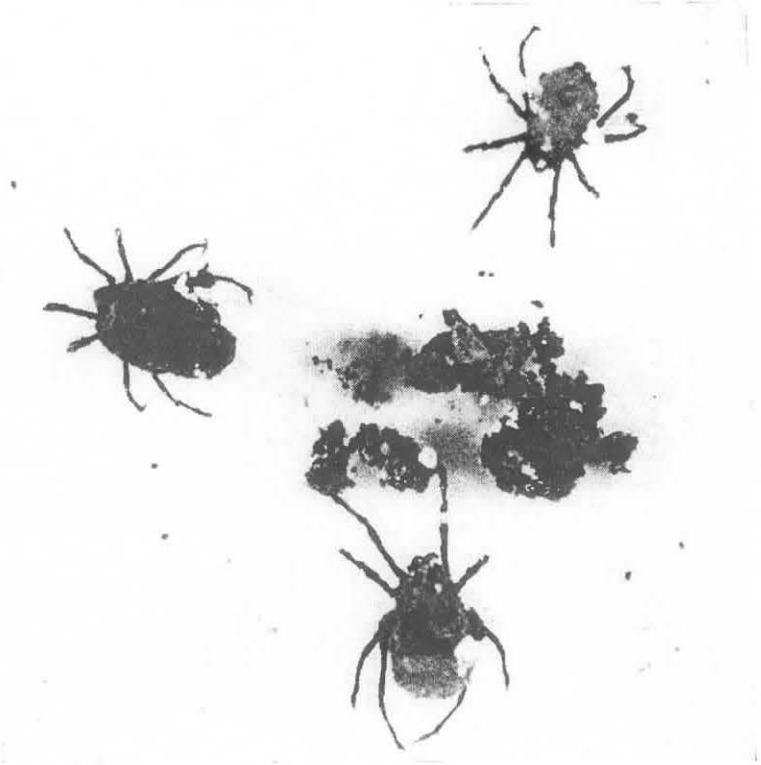

PLATE 2.

The juvenile stages of *B. geniculosa*. The debris covering the integument has been removed from two of the nymphs (right), exposing the soft, non-sclerotized cuticle (× 10).

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